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Letter to the Editor

THE EFFECTS OF AMMONIA ON FRESHWATER UNIONID MUSSELS

To the Editor:

Freshwater mussels (Family Unionidae) are ecologically and economically important in aquatic ecosystems. Unionids often comprise a significant proportion of the total biomass in freshwater benthic communities, are important in nutrient cycling, and mix surficial sediments through bioturbation. Unionids also serve as food for many mammals, including raccoons, muskrats, and otters. The density and species diversity of unionids in North America have declined substantially during the past century, but the causal factors are largely unknown. Suggested causes have included sedimentation, changes in fish–host distribution, impoundment of rivers, and introduction of exotic species. Although not sufficiently documented, exposure to toxic contaminants also may have contributed to these declines. Chemical spills and other point sources of contaminants have caused localized mortality; however, widespread decreases in density and diversity more likely have resulted from the subtle, pervasive effects of chronic, low-level contamination [1].

Certain natural-history characteristics make unionids useful monitors of water and sediment pollution. Adults are macroscopic, long-lived (30–130 years), infaunal benthic invertebrates that obtain food principally by filter-feeding and are consequently exposed to contaminants that have been dissolved in water, associated with suspended particles, and deposited in bottom sediments. Their association with fine-grained sediments increases their exposure to sediment-associated contaminants. Recent research suggests that at least a portion of the juvenile life-history stage in some species involves feeding primarily on particles associated with sediments and pore water. Recent research also suggests that at least some species of juvenile unionids feed primarily on particles associated with sediments and pore water during their early development [2]. This has important implications for ecotoxicological studies, because sediments adsorb many contaminants that partition into pore water. Unfortunately, few toxicological data are available for unionids. Furthermore, the bulk of the unionid toxicity literature is based on water-only exposures, even though several studies have shown that sediment-associated contaminants likely contributed to the decline of mollusks in several large rivers [3,4]. Unionid ecotoxicology could benefit from the approaches used in the marine bivalve literature to help ascertain the importance of different routes of contaminant exposure (surface water, pore water, sediments, and food). Although unionids have been used for decades as indicators of contaminant bioavailability and exposure, the effects of contaminants on unionids are largely unknown. Ultimately, we need to link bioavailability and exposure to adverse effects.

The field of molluskan ecotoxicology largely began with studies of fingernail clams, which are organisms in the same class (Bivalvia) as unionids but whose substantial differences in reproduction and life span generally preclude their use as surrogates for unionids. During the late 1980s, it became apparent that relative to other invertebrates and fishes, unionids were more sensitive to certain contaminants. Consequently, there has been substantial interest in standardizing toxicity tests with unionids. Conducting toxicity tests with unionids, however, poses challenges not routinely encountered with more traditional test species (i.e., *Daphnia*, *Pimephales*). These challenges include maintaining test organisms in suitable physiological health before and during testing, choosing exposure durations representative of their long life span, and identifying suitable response endpoints. For example, growth and reproduction, two commonly used endpoints in toxicological evaluations of other aquatic invertebrates, have limited application in short-term tests with unionids. Growth of adult unionids is not an appropriate endpoint because of their long life span and slow growth rates, although recent data suggest this may be a sensitive endpoint in juveniles [5]. Reproductive endpoints are difficult to measure, because most unionids are not sexually dimorphic and females may not reproduce annually. Although juveniles are generally more sensitive than larvae or adults to contaminants, culturing juveniles is time-consuming and possible only in approximately 50% of the nearly 300 species for which fish hosts have been identified. Additionally, the small size of juveniles (50–400 μg) may not provide sufficient mass for certain analyses and endpoints. However, the field has made great strides in the past decade, and many of these challenges are being addressed.

Ammonia is an important contaminant in aquatic environments because it is highly toxic and ubiquitous in surface waters [6]. Sources of ammonia include industrial, municipal, and agricultural wastewaters as well as precipitation and natural processes, such as decomposition of organic nitrogen. Recent evidence suggests that atmospheric deposition is one of the most rapidly growing sources of anthropogenic nitrogen entering aquatic environments, and domestic livestock are the largest documented global source of atmospheric ammonia [7]. Total ammonia nitrogen is the sum of two forms in equilibrium: the ammonium ion (NH_4^+) and un-ionized ammonia (NH_3). The toxicity of ammonia to aquatic organisms is generally attributed to NH_3, although NH_4^+ can contribute significantly to toxicity under certain conditions. In freshwater, the percentage of total ammonia in the NH_3 form depends primarily on pH and temperature; as temperature and pH increase, the percentage of total ammonia as NH_3 increases.

The recent case of hypoxia in the Gulf of Mexico has generated substantial interest regarding nitrogen dynamics in large rivers. Data from the hypoxia studies suggest that surficial sediments are enriched with nitrogen in several systems in which populations of benthic invertebrates, including unionids, have declined. Unionids may be relatively insensitive to some pesticides and organic compounds, but their early life-history stages are among the most sensitive of aquatic fauna tested for metals, chlorine, and NH_3. For example, 96-h median lethal concentrations (LC50s) in four species of juvenile unionids range from 40 to 280 μg NH_3/L [5,8,9]; similar 96-h LC50s for more traditional test species range from 1,830 to 2,550 μg NH_3/L in *Pimephlaes promelas* [10], from 190 to 6,090 μg NH_3/L in *Hyalella azteca* [11], and from 260 to 1,040 μg NH_3/L in *Oncorhynchus mykiss* [10]. Although only a fraction of unionid species have been tested thus far, the available data suggest that juveniles are quite sensitive to NH_3. In addition, when expressed as total ammonia nitrogen and normalized to pH 8, the LC50s for juvenile unionids are substantially less than the acute national water-quality criterion [5,12], suggesting that existing criteria may not be protecting unionids. The latest update of the water-quality criteria in the United States made the ammonia standard less stringent under certain environmental conditions [13].

The following publications discuss the sensitivity of juvenile unionids to ammonia and their potential utility as toxicity test organisms. Mummert et al. [9] report on the effects of waterborne ammonia on survival of juvenile unionids in the laboratory. Newton et al. [5] report on the effects of ammonia in pore water on survival and growth of juvenile unionids in laboratory tests, and Bartsch et al. [14] examine the role of juvenile unionids as in situ biomonitors for ammonia in the field. Finally, Augspurger et al. [12] estimate ammonia concentrations that would be protective of unionids and compare these to the recently revised water-quality criteria for ammonia. In total, these publications show that early life-history stages of unionids are quite sensitive to NH_3. These papers represent a major advancement in the field of ecotoxicological studies with unionids and a common, but toxic, contaminant. If water-quality criteria become more stringent and pollution-prevention controls more effective, point-source pollution likely will have a decreasing influence on water and sediment quality. The future challenges in unionid ecotoxicology will be refining test methods to better address chronic exposures and sublethal effects, establishing causal linkages to integrate laboratory and field observations, and measuring the cumulative effects of long-term exposure to nonpoint and multiple stressors.

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